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SOIL INVERTEBRATE AND MICROBIAL POPULATIONS UNDER THREE TREE SPECIES ON THE SAME SOIL TYPE

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ABSTRACT.—The surface mineral soil beneath an aspen stand contained about 10 times as many bacteria (*Corynebacteria, Mycobacteria*, and *Nocardia*) and 30 to 50 percent more fungi (*Trichoderma, Aspergillus, Cephalosporium*, and *Fusarium*) than did soil beneath two conifer stands. These organisms were 10 to 1,000 times more abundant in the surface 10 cm than in the next 15 cm. Red pine had more annelids and fewer arachnids in the forest floor than did white spruce and quaking aspen.

KEY WORDS: Bacteria, fungi, annelida, archnida, decomposer organisms.

Nutrient cycles in forest ecosystems are highly complex. Contributing to this complexity is the critical role microorganisms and invertebrates play in the breakdown of organic matter, and in nutrient mineralization and immobilization. Soil organisms are affected by temperature, moisture, pH, nutrients, aeration, and by litter quality (ease of breakdown), all of which are influenced by plant species composition (Swift *et al*. 1979). In a study of 39-year-old adjacent stands of red pine, (*Pinus resinosa* Ait.), white spruce (*Picea glauca* (Moench) Voss), and quaking aspen (*Populus tremuloides* Michx.) on two soils, Perala and Alban (1982a) found that quaking aspen had less litterfall than red pine or white spruce, but higher litterfall nutrient levels for phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg). Aspen also had the lowest organic matter and carbon content (Alban 1982), but it is not clear whether this was due to the lower litterfall, more rapid litter decomposition, or both.

Little is known about the differences in soil organism populations that may exist between different forest stands, or how those differences may be important. As a first step, this note reports on a survey of species composition and population density of microorganisms and soil invertebrates in forest floor and upper mineral soil for red pine, white spruce, and quaking aspen stands growing adjacent to one another on the same soil type.

THE STUDY SITE

The stands have been intensively studied for distribution of biomass, organic matter, and nutrients (Alban *et al*. 1978; Alban 1982; Perala and Alban 1982a, 1982b) and are located on the Pike Bay Experimental Forest in north central Minnesota (47° 21′N, 94° 30′W). The stands all originated after clearcut logging in 1933. The aspen regenerated by suckering, and the pine and spruce by planting in the fall of 1934. The stands (table 1) are unthinned and all are in 0.4 ha blocks.

Table 1.—*Characteristics of three 39-year-old forest stands*

Stand	Trees per hectare	Mean height	Mean d.b.h. (outside bark)	Basal area	Volume inside bark	Site index (50 years)
	Number	*m*	*cm*	------- m^2/ha -------		*m*
Aspen						
Aspen	1,334	20.3	18	34.7	286	22.9
Other species	1,655	11.0	7	7.0	39	—
White spruce	2,187	14.4	15	41.1	256	18.3
Red pine	1,780	17.6	19	51.9	408	20.7

Climate is continental, meaning cold winters (mean January temperature −14°C), cool summers (mean July temperature 20°C), and 610 mm of precipitation (half falls during the growing season).

The soil has developed from calcareous glacial till and is classified as Warba very fine sandy loam (Glossic Eutroboralf); it is well drained and moderately acid (pH 5-6) above the C horizon. The soil occurs on a gently undulating till plain and is morphologically uniform for the entire study area.

METHODS

In August 1982, we collected three samples of forest floor and mineral soil (0-10 cm, and 10-25 cm) from under each stand, using a cylindrical 105 cm^2 soil sampling tool (Jurgensen *et al*. 1977). The three 0-10 cm mineral soil samples for each stand were kept separate; the forest floor and 10-25 cm depth samples were composited to give one sample for each stand. Mineral soil samples were stored at 4°C until analyzed.

Microbial Analysis

We determined soil microbial populations by the agar-plate culture method. Immediately before analysis, each sample was passed through a sterilized 10-mesh soil sieve, mixed thoroughly, and subsampled to determine ovendry weight and moisture content (105°C).

For each of the mineral soil collections, we established a dilution series of 10^{-1} to 10^{-7} for plating the soil by starting with 10 grams of soil in 90 ml of sterilized water, agitating vigorously for 10 minutes, and transferring 10 ml of the soil suspension by a sterilized pipette to a 90 ml water blank. Successive transfers were made to provide all dilutions for each sample.

From each of the seven dilutions prepared for each sample, 1 ml of soil suspension was added by sterile pipette to each of five sterile petri dishes. About 12 ml of agar (24°C) was added to each plate and carefully swirled to ensure even distribution. Agar was allowed to solidify, the plate was placed inverted in a dark 27°C incubator, and left undisturbed.

After 14 days, we recorded sample numbers of bacteria and fungal colonies. Dilution plates with intermediate numbers of colonies and growth provided the most reliable data.

Analyses of variance were conducted after log (N + 1) transformation. Means were then retransformed to arithmetic values and multiplied by the appropriate dilution factors and soil moisture content to express counts on a dry soil basis.

Different colonial types of bacteria and fungi were subcultured and subsequently examined. Bacteria were classified as either gram-positive or gram-negative. We performed the acid-fast stain to identify to genus level (Norris and Ribbons 1971). Fungal types were determined from the plate colonies by slide preparation and by direct plate observations (Barnett 1962).

Invertebrate Analysis

Worms and larger arthropods were hand-sorted from the moist forest floor samples. We extracted smaller insects after carefully dispersing sample material over a white sheet of paper.

RESULTS

Bacteria—Actinomycetes

In the 0-10 cm layer, there were about 10 times as many bacteria under aspen than under the conifers (table 2). In the 10-25 cm layer, there were 8 to 12 times as many bacteria under red pine than under the other tree species. These counts were much lower than in the 0-10 cm layer. Seventy-five percent of all bacteria under the quaking aspen stands were gram-positive "acid-fast," short-rods. These properties are

Table 2.—*Number of bacterial colonies per gram of dry soil for three forest stands*[1]

Stand	Soil strata	
	0-10 cm	10-25 cm
	Number of colonies (× 10^4)	
Red pine	56a	5.40
White spruce	50a	.46
Quaking aspen	640b	.67

[1]Means followed by the same letter do not differ significantly at the 0.5 percent level.

characteristic of the genera *Corynebacteria, Mycobacteria*, and *Nocardia*. The bacteria under red pine and white spruce were predominantly gram-positive coccoid rods—possibly actinomycetes.

Fungi

In the 0-10 cm layer, soil fungi were significantly more numerous under aspen than under conifers (table 3). Soil fungi in the 10-25 cm layer were about one-tenth as numerous as in the 0-10 cm layer.

A penicillium-like fungus was the most predominant soil fungus present; *Trichoderma, Aspergillus, Cephalosporium*, and *Fusarium* comprised the main genera.

Large Invertebrates

Soil invertebrate populations were about equal under all species (table 4). In red pine, the soil invertebrate population was composed of 45 percent worms, 40 percent insects (beetles, larvae, and springtails), but no arachnids (mites and spiders). The remaining 15 percent of the population was not identifiable. The white spruce and quaking aspen stands had similar invertebrate populations, but with fewer worms and more arachnids than the red pine.

Table 3.—*Number of fungal colonies per gram of dry soil for three forest stands*[1]

Stand	Soil strata	
	0-10 cm	10-25 cm
	Number of colonies (× 10^4)	
Red pine	4.6a	0.42
White spruce	3.9a	.35
Quaking aspen	6.0b	.37

[1]Means followed by the same letter do not differ significantly at the 1 percent level.

Table 4.—*Invertebrates in the forest floor* (In number per m^2)

	Red pine	White spruce	Quaking aspen
Annelida—worms	726	444	403
Insects—beetles beetle larvae springtails	645	847	1,250
Arachnids—mites Spiders	0	323	282
Unidentified—	242	323	282
Total	1,613	1,937	2,217

DISCUSSION

The plate-count method for total microbial count is a highly selective procedure, depending on the nutritional, chemical, and physical conditions employed (Parkinson *et al.* 1971). Nevertheless, the very large bacterial differences between tree species and the statistically significant fungal differences in the 0-10 cm layer indicate that different tree species provide different surface soil environments for microorganisms.

The reasons for the greater microbial populations under the aspen are not apparent, but some common assumptions appear to be invalid. It is often stated (Russell 1973) that bacteria are favored over fungi at higher pH's. In these stands, however, the most bacteria and fungi occur under aspen with a surface soil pH of 5.6, and the least under spruce with a very similar pH of 5.4 (*cf.* tables 2, 3, and 5). Conversely, bacteria and fungi do not differ between red pine and white spruce, even though the pH of the red pine surface soil is 0.6 units greater than that of the spruce. Clearly, microorganism counts are not closely related to soil pH.

Table 5.—*Organic matter and pH of surface mineral soils under three forest stands (Alban 1982)*[1]

Stand	Organic matter		pH	
	0-10 cm	10-25 cm	0-10 cm	10-25 cm
	----- *t/ha* -----			
Red pine	58a	15a	6.0a	5.9a
White spruce	49a	16a	5.4b	5.5b
Quaking aspen	35b	10b	5.6b	5.9a

[1]Means followed by the same letter do not differ significantly at the 0.05 level.

There was significantly less organic matter in the surface soil under aspen than under conifers (table 5). This is consistent with the larger number of bacteria and fungi under aspen, which would lead to faster decomposition of organic matter under the aspen.

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